Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
L2	113	EDTA same europium	USPAT	OR	OFF	2004/12/28 17:17
L3	44	EDTA same europium same chelate	USPAT	OR	OFF	2004/12/28 17:20
L4	469	delmas.in.	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:21
L5	10	l4 and (fracture or osteoporosis or bone)	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:21
L6	1	l5 and antibody and 17-24	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:22
L7	4	osteocalcin and antibody and EDTA and 17-24	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:24
L8	751	osteocalcin and antibody and EDTA	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:24
L9	0	l8 and (antibody same 17-24)	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:25
L10	4	l8 and 17-24	US-PGPUB; USPAT; EPO; DERWENT	OR	OFF	2004/12/28 17:25

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 NEWS  2          "Ask CAS" for self-help around the clock
 NEWS  3  SEP 01  New pricing for the Save Answers for SciFinder Wizard within
                  STN Express with Discover!
 NEWS  4  OCT 28  KOREAPAT now available on STN
 NEWS  5  NOV 30  PHAR reloaded with additional data
 NEWS  6  DEC 01  LISA now available on STN
 NEWS  7  DEC 09  12 databases to be removed from STN on December 31, 2004
 NEWS  8  DEC 15  MEDLINE update schedule for December 2004
 NEWS  9  DEC 17  ELCOM reloaded; updating to resume; current-awareness
                  alerts (SDIs) affected
 NEWS  10 DEC 17  COMPUAB reloaded; updating to resume; current-awareness
                  alerts (SDIs) affected
 NEWS  11 DEC 17  SOLIDSTATE reloaded; updating to resume; current-awareness
                  alerts (SDIs) affected
 NEWS  12 DEC 17  CERAB reloaded; updating to resume; current-awareness
                  alerts (SDIs) affected
 NEWS  13 DEC 17  THREE NEW FIELDS ADDED TO IFIPAT/IFIUDB/IFICDB

 NEWS EXPRESS  OCTOBER 29 CURRENT WINDOWS VERSION IS V7.01A, CURRENT
               MACINTOSH VERSION IS V6.0c(ENG) AND V6.0Jc(JP),
               AND CURRENT DISCOVER FILE IS DATED 11 AUGUST 2004
 NEWS HOURS    STN Operating Hours Plus Help Desk Availability
 NEWS INTER    General Internet Information
 NEWS LOGIN    Welcome Banner and News Items
 NEWS PHONE    Direct Dial and Telecommunication Network Access to STN
 NEWS WWW      CAS World Wide Web Site (general information)
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=> osteocalcin and antibody and (bivalent or Ca or EDTA)
L1            1 FILE AGRICOLA
L2            9 FILE BIOTECHNO
L3            1 FILE CONFSCI
L4            0 FILE HEALSAFE
L5            0 FILE IMSDRUGCONF
L6            5 FILE LIFESCI
L7            0 FILE MEDICONF
L8            6 FILE PASCAL

TOTAL FOR ALL FILES
L9           22 OSTEOCALCIN AND ANTIBODY AND (BIVALENT OR CA OR EDTA)

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DUPLICATE IS NOT AVAILABLE IN 'IMSDRUGCONF, MEDICONF'.
ANSWERS FROM THESE FILES WILL BE CONSIDERED UNIQUE
PROCESSING COMPLETED FOR L9
L10           15 DUP REM L9 (7 DUPLICATES REMOVED)

=> d l10 ibib abs total

L10  ANSWER 1 OF 15  LIFESCI    COPYRIGHT 2004 CSA on STN DUPLICATE 1
ACCESSION NUMBER:   2004:1808  LIFESCI
TITLE:              Immunohistochemical examination on intracranial
                    calcification in neurodegenerative diseases
AUTHOR:             Fujita, Daisuke; Terada, Seishi; Ishizu, Hideki; Yokota,
                    Osamu; Nakashima, Hanae; Ishihara, Takeshi; Kuroda,
                    Shigetoshi
CORPORATE SOURCE:   Department of Neuropsychiatry, Okayama University Graduate
                    School of Medicine and Dentistry, 2-5-1 Shikata-cho,
                    Okayama 700-8558, Japan
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SOURCE: Acta Neuropathologica [Acta Neuropathol.], (20030300) vol. 105, no. 3, pp. 259-264.
ISSN: 0001-6322.
DOCUMENT TYPE: Journal
FILE SEGMENT: N3
LANGUAGE: English
SUMMARY LANGUAGE: English

AB Fahr-type calcification is a relatively common finding in the elderly, and in younger patients with Alzheimer's disease, calcification in the basal ganglia is not uncommon. However, as far as we know, an immunohistochemical study of intracranial calcification in neurodegenerative diseases has not been performed. In this study, we examined intracranial calcification of the basal ganglia and cerebellum with **antibodies** against noncollagenous bone matrix proteins. Nineteen brains were employed. The diagnoses were diffuse neurofibrillary tangles with calcification in five, Alzheimer's disease in five, Pick's disease in one, progressive supranuclear palsy in one, Parkinson's disease in one, and six controls. By conventional histology, three patterns of calcium (**Ca**) deposition were recognized: diffuse deposition within the tunica media of small and medium-sized vessels (type 1 deposition), free spherical or lobulated concretions (type 2 deposition) in the parenchyma, and rows of small calcospherites lying along capillaries (type 3 deposition). Type 3 deposition is relatively rare, and may be a hallmark of severe intracranial calcification. Immunohistochemistry demonstrated that osteopontin was present diffusely in all **Ca** deposition types. **Osteocalcin** was present chiefly in the peripheral region of type 2 and 3 depositions, as well as in only the rims of type 1 deposition. Bone sialoprotein and osteonectin were found only in the core portions of type 2 and 3 depositions. In brief, type 1 deposition shows a different staining pattern from type 2 and 3. Different **Ca** deposition patterns of noncollagenous bone matrix proteins may suggest their separate roles in the pathogenesis of intracranial calcification.

L10 ANSWER 2 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
ACCESSION NUMBER: 2000:32876396 BIOTECHNO
TITLE: Slow rates of degradation of **osteocalcin**: Green light for fossil bone protein?
AUTHOR: Collins M.J.; Gernaey A.M.; Nielsen-Marsh C.M.; Vermeer C.; Westbroek P.
CORPORATE SOURCE: M.J. Collins, Fossil Fuels Environ. Geochemistry, Postgraduate Institute, Newcastle Research Group, Newcastle upon Tyne, NE1 7RU, United Kingdom.
E-mail: m.collins@ncl.ac.uk
SOURCE: Geology, (2000), 28/12 (1139-1142), 37 reference(s)
CODEN: GLGYBA ISSN: 0091-7613
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English

AN 2000:32876396 BIOTECHNO
AB Our claim, published in this journal, for successful immunodetection of the protein **osteocalcin** in dinosaur bone has been challenged on the grounds that the findings are inconsistent with the kinetics of decomposition. Here we show that the close association of **osteocalcin** to the bone mineral vastly enhances its preservation potential relative to the same protein in aqueous solution. We conducted heating experiments (75-95 °C) of modern bone powder and monitored the survival of three different regions of **osteocalcin** (N-terminal, His.sub.4-Hyp.sub.9; C-terminal, Phe.sub.4.sub.5-Val.sub.4.sub.9; and the mid-region, Pro.sub.1.sub.5-Glu.sub.3.sub.1) with monoclonal **antibodies**. Extrapolation of our data to 10 °C ambient burial temperatures indicates that preservation of the γ-carboxylated mid-region in fossil bone cannot be excluded on

kinetic grounds. Clearly, in situ sequence analysis will be the only method by which the preservation of fossil macromolecules will be unequivocally established. Nevertheless, our findings demonstrate the importance of mineral association to protein survival, as was borne out by an investigation of Holocene (**ca**. 6 ka) bones. Only in those samples with little recrystallization was the γ-carboxylated mid-region well preserved. These results imply that the future success of ancient biomolecule research largely depends on our understanding the interaction between these materials and their environment throughout diagenesis.

L10 ANSWER 3 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
DUPLICATE
ACCESSION NUMBER: 2000:30152258 BIOTECHNO
TITLE: Development and evaluation of three immunofluorometric assays that measure different forms of **osteocalcin** in serum
AUTHOR: Kakonen S.-M.; Hellman J.; Karp M.; Laaksonen P.; Obrant K.J.; Vaananen H.K.; Lovgren T.; Pettersson K.
CORPORATE SOURCE: S.-M. Kakonen, Univ. of Texas Health Science Center, Department of Medicine, Division of Endocrinology, 7703 Floyd Curl Dr., San Antonio, TX 78284-7877, United States.
E-mail: kakonen@uthscsa.edu
SOURCE: Clinical Chemistry, (2000), 46/3 (332-337), 33 reference(s)
CODEN: CLCHAU ISSN: 0009-9147
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 2000:30152258 BIOTECHNO
AB Background: Circulating human **osteocalcin** (hOC) has been used as a marker of bone formation. Our aim was to validate three immunofluorometric assays (IFMAs), measuring different forms of hOC. Methods: The two-site IFMAs were based on previously characterized monoclonal **antibodies**. Assay 2 recognized intact hOC, assays 4 and 9 measured the NH.sub.2-terminal mid-fragment and the intact hOC. In addition, assay 9 required hOC to be γ-carboxylated. Results: A 76-79% increase of serum immunoreactive hOC was found in the postmenopausal group compared with the premenopausal group with all IFMAs. With **EDTA**-plasma samples, the observed increases were lower (49-65%). The hOC concentration in the postmenopausal group receiving hormone replacement therapy was 42-44% lower than that in the postmenopausal control group in both serum and **EDTA**-plasma samples. The depressed carboxylation in warfarin- treated patients was accompanied by lower results in assay 9. The ratio of assay 9 to assay 4 totally discriminated the warfarin-treated patients from the controls. Assay 9 showed the smallest decreases in measured hOC after storage of serum or plasma for 4 weeks at 4 °C, followed by assay 4 and assay 2. Results from the last assay were <17% of their initial values after 4 weeks of storage. No diurnal variation was observed with assay 9 as opposed to the two other IFMAs. Conclusion: The three assays with their distinct specificity profiles (intact vs fragmented and carboxylated vs decarboxylated hOC) may provide valuable tools for investigating the significance of different hOC forms in various bone-related diseases. (C) 2000 American Association for Clinical Chemistry.

L10 ANSWER 4 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
ACCESSION NUMBER: 2000:30097339 BIOTECHNO
TITLE: The effect of bone morphogenetic protein-7 on the expression of type I inositol 1,4,5-trisphosphate receptor in G-292 osteosarcoma cells and primary osteoblast cultures

AUTHOR: Bradford P.G.; Maglich J.M.; Ponticelli A.S.; Kirkwood K.L.
CORPORATE SOURCE: P.G. Bradford, Dept. of Pharmacology and Toxicology, Sch. of Med. and Biomedical Sciences, State University of New York, Buffalo, NY 14214-3000, United States. E-mail: pgb@acsu.buffalo.edu
SOURCE: Archives of Oral Biology, (2000), 45/2 (159-166), 32 reference(s)
CODEN: AOBIAR ISSN: 0003-9969
PUBLISHER ITEM IDENT.: S0003996999001223
DOCUMENT TYPE: Journal; Article
COUNTRY: United Kingdom
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 2000:30097339 BIOTECHNO
AB Bone morphogenetic protein-7 (BMP-7) affects differentiation of preosteoblasts enabling the resultant cells to respond optimally to acutely acting regulators. As the phosphoinositide cascade and, particularly, the calcium-mobilizing inositol 1,4,5-trisphosphate (InsP.sub.3) receptor are integral to stimulus-secretion coupling in osteoblasts, the hypothesis that BMP-7 affects InsP.sub.3 receptor expression was examined in the G-292 human osteosarcoma cell line and in primary cultures of human osteoblasts. G-292 osteosarcoma cells were found to be a valid experimental model for primary human osteoblasts, expressing osteoblastic mRNAs encoding **osteocalcin**, bone sialoprotein, alkaline phosphatase, α1-collagen, epidermal growth-factor receptor, and BMP type II receptor. When cultured long term in the presence of ascorbic acid and β-glycerophosphate, G-292 cells underwent further osteoblastic differentiation, forming nodules and exhibiting restricted mineralization. G-292 cells responded to BMP-7 with an increase in InsP.sub.3 receptor density. Ligand-binding studies established that BMP-7 (50 ng/ml) treatment of G-292 cells increased InsP.sub.3 receptor density 2.4-fold with no apparent change in affinity. Immunoblot analysis with **antibodies** specific for type I, type II, and type III InsP.sub.3 receptors revealed that BMP-7 (50 ng/ml) treatment resulted in a specific increase (206 ± 8%) in the type I receptor. Reverse transcription-polymerase chain reaction and Northern blot analyses of G-292 and primary human osteoblasts confirmed an increase in type I InsP.sub.3 receptor mRNA upon BMP-7 treatment. These results demonstrate that G-292 cells respond to BMP-7 with an increase InsP.sub.3 receptor density, consistent with the enhanced capacity of these cells to respond to **Ca**.sup.2.sup.+-mobilizing secretory hormones during osteoblast differentiation.

L10 ANSWER 5 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
DUPLICATE
ACCESSION NUMBER: 1999:29165963 BIOTECHNO
TITLE: Immunoradiometric assay for intact human **osteocalcin**(I-49) without cross-reactivity to breakdown products
AUTHOR: Colford J.W.; Lueddecke B.A.; Salvati M.; Hanna D.; Sailer D.; Khosla S.; Riggs B.L.; Langman C.B.
CORPORATE SOURCE: C.B. Langman, DiaSorin Inc., 1990 Industrial Blvd., Stillwater, MN, United States.
SOURCE: Clinical Chemistry, (1999), 45/4 (526-531), 21 reference(s)
CODEN: CLCHAU ISSN: 0009-9147
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 1999:29165963 BIOTECHNO
AB Background: **Osteocalcin** (Oc), a serum marker of bone turnover, circulates in several forms. We developed an assay for intact human Oc

and investigated its clinical features. Methods: We generated goat **antibodies** and N- and C-terminal Oc. The former was used on solid phase (polystyrene beads), and the latter was used as the tracer in an IRMA. Results: The assay was linear with no Cross-reactivity to Oc(1,43), total imprecision (CV) of <10%, and recovery of 100% ± 10%. Assay values for intact Oc in **EDTA** plasma samples were unchanged at 18-25 °C for 6 h. Values for intact Oc in serum, **EDTA** plasma, and heparin plasma samples did not change after storage on ice for 8 h. Serum samples from patients with various conditions were stored at - 70 or -135 °C for up to 5 years and yielded z-scores comparable to an Oc(1- 43) IRMA for all conditions except for renal failure. In renal failure, the Oc(1-43) assay values were increased, whereas the intact assay values were in the reference interval. Conclusion: Decreases in Oc assay values are inhibited by calcium chelation, and slowed by reduced temperatures. The described assay for intact Oc allows improved specificity for bone compared with an assay for Oc(1-43).

L10 ANSWER 6 OF 15 LIFESCI COPYRIGHT 2004 CSA on STN
ACCESSION NUMBER: 1998:113268 LIFESCI
TITLE: A novel human bone marrow stroma-derived cell line TF274 is highly osteogenic in vitro and in vivo
AUTHOR: Prabhakar, U.; James, I.E.; Dodds, R.A.; Lee-Rykaczewski, E.; Rieman, D.J.; Lipshutz, D.; Trulli, S.; Jonak, Z.; Tan, K.B.; Drake, F.H.; Gowen, M.
CORPORATE SOURCE: Clinical Pharmacology Unit, 51 N, 39th St., Presbyterian Medical Center, Philadelphia, PA 19406, USA
SOURCE: Calcif. Tissue Int., (19980900) vol. 63, no. 3, pp. 214-220
ISSN: 0171-967X.
DOCUMENT TYPE: Journal
FILE SEGMENT: T
LANGUAGE: English
SUMMARY LANGUAGE: English
AB A novel, immortalized, human bone marrow stroma-derived cell line TF274 is described which has the ability to form bone both in vitro and in vivo. Under basal conditions these cells expressed alkaline phosphatase (ALP) and type I collagen genes which are characteristic of the osteoblast phenotype. ALP levels were upregulated in the presence of osteotropic agents such as parathyroid hormone (PTH), transforming growth factor beta (TGF- beta), and BMP-2. In addition, PTH also increased cAMP levels in these cells. The capacity of these cells to form bone in vitro was evaluated by culturing them in the presence of L-ascorbic acid and beta -glycerophosphate. Matrix mineralization in these cultures was assessed by Alizarin Red staining and increased super(45)**Ca** uptake. Under these conditions mineralized nodule formation was observed in less than 2 weeks. Northern analysis of TF274 cells at various times during the mineralization process indicated a temporal expression of the **osteocalcin** gene that is typically associated with differentiating osteoblasts. The osteogenic nature of TF274 cells was confirmed in vivo using the severe combined immunodeficient (SCID) mouse model. **Antibodies** to human leukocyte antigens (HLA), class I antigens, and human OK super(a) blood group antigen were used to demonstrate that the lesions formed were of human origin. By 21 days, the lesion consisted of a homogeneous focus of ALP-positive cells containing areas of mineralized bone lined with tartarate-resistant acid phosphatase (TRAP) positive osteoclasts. Thus, the TF274 cells exhibit osteogenic potential both in vitro and in vivo. This immortalized cell line represents a consistent source of cells that can be used to study human osteoblast differentiation both in vitro and in vivo.

L10 ANSWER 7 OF 15 AGRICOLA Compiled and distributed by the National Agricultural Library of the Department of Agriculture of the United States of America. It contains copyrighted materials. All rights reserved.

(2004) on STN DUPLICATE 4

ACCESSION NUMBER: 97:12365 AGRICOLA
DOCUMENT NUMBER: IND20546934
TITLE: The determination of serum concentrations of **osteocalcin** in growing pigs and its relationship to end-measures of bone mineralization.
AUTHOR(S): Carter, S.D.; Cromwell, G.L.; Combs, T.R.; Colombo, G.; Fanti, P.
CORPORATE SOURCE: North Dakota State University, Fargo.
SOURCE: Journal of animal science, Nov 1996. Vol. 74, No. 11. p. 2719-2729
Publisher: Champaign, Ill. : American Society of Animal Science.
CODEN: JANSAG; ISSN: 0021-8812
NOTE: Includes references
PUB. COUNTRY: Illinois; United States
DOCUMENT TYPE: Article
FILE SEGMENT: U.S. Imprints not USDA, Experiment or Extension
LANGUAGE: English

AB **Osteocalcin**, a 49-amino acid, gamma-carboxyglutamic acid-containing protein produced by the osteoblast, has been shown in laboratory animals to be a better marker of bone turnover than alkaline phosphatase. To determine serum **osteocalcin** levels in growing pigs, we isolated pure porcine **osteocalcin** and developed a double-**antibody** RIA. To evaluate the effects of dietary **Ca** and P levels on serum **osteocalcin**, 36 individually penned crossbred pigs (19.5 kg initial BW) were fed fortified corn-soybean meal diets (.95% lysine) containing four levels of **Ca** (.42, .66, .90, 1.14%) and P (.35, .55, .75, .95%) in a 30-d test. Increasing dietary **Ca** and P improved body weight gain quadratically ($P < .02$). Most bone traits improved quadratically ($P < .05$) with increasing **Ca** and P. Pigs were bled on d 0, 10, 20, and 30 to determine serum levels of alkaline phosphatase, 1,25-dihydroxyvitamin D3, and **osteocalcin**. **Osteocalcin** decreased ($P < .02$) linearly with increasing **Ca** and P on d 10, 20, and 30. However, this effect was much more pronounced on d 20 and 30. Alkaline phosphatase decreased with the first incremental increase in dietary **Ca** and P, but was not affected by higher levels on any day measured. **Osteocalcin** was inversely correlated with growth rate ($r = -.54$, $P < .01$), bone strength ($r = -.57$, $P < .01$), metacarpal ash ($r = -.29$, $P < .10$), femur ash ($r = -.60$, $P < .01$), and femur ash weight ($r = -.65$, $P < .01$). Similar results were found for 1,25-dihydroxyvitamin D3. Alkaline phosphatase was not correlated with performance or most bone traits on d 30. Based on this model, these results suggest that serum **osteocalcin** and 1,25-dihydroxyvitamin D3 are better predictors of bone mineralization and (or) turnover in pigs than serum alkaline phosphatase.

L10 ANSWER 8 OF 15 PASCAL
on STN

ACCESSION NUMBER: 1997-0077373 PASCAL
COPYRIGHT NOTICE:
TITLE (IN ENGLISH): Acquired osteosclerosis associated with intravenous drug use and hepatitis C infection
AUTHOR: DIAMOND T.; DEPCZYNSKI B.
CORPORATE SOURCE: Department of Endocrinology, St. George Hospital, Kogarah, NSW, Australia; Department of Endocrinology, St. Vincent's Hospital, Darlinghurst, NSW, Australia
SOURCE: Bone : (New York), (1996), 19(6), 679-683, 21 refs.
ISSN: 8756-3282
DOCUMENT TYPE: Journal; (case report, clinical case)
BIBLIOGRAPHIC LEVEL: Analytic
COUNTRY: United States
LANGUAGE: English

AVAILABILITY: INIST-19041, 354000061214540170
AN 1997-0077373 PASCAL

AB Hepatitis C has recently been recognized as a secondary cause of osteosclerosis ; a further example, the first outside of North America, is described. A 37-year-old man with a history of intravenous drug use and known to be hepatitis C **antibody** positive presented with bone pain. Radiographs and magnetic resonance imaging demonstrated an increase in cortical and trabecular bone that on biopsy was of a normal lamellar pattern but markedly sclerotic. Biochemical markers of bone formation (serum **osteocalcin**) and resorption (urinary hydroxyproline excretion rate) were both markedly elevated. Pain lessened following administration of pamidronate. Biochemical markers of bone turnover fell towards their reference ranges 12 months after initiating pamidronate therapy but without significant change in bone mineral density. Osteosclerosis is a rare complication of hepatitis C infection, the symptoms of which are controllable with diphosphonate therapy.

L10 ANSWER 9 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
DUPLICATE
ACCESSION NUMBER: 1993:23157799 BIOTECHNO
TITLE: Development and characterization of a polyclonal antiserum-based radioimmunoassay for dog **osteocalcin**
AUTHOR: Fanti P.; Colombo G.; Yao C.; Brown S.A.; Vernon M.W.; Malluche H.H.
CORPORATE SOURCE: Division of Nephrology, Kentucky University Medical Center,Lexington, KY 40536, United States.
SOURCE: Journal of Bone and Mineral Research, (1993), 8/6 (745-752)
CODEN: JBMREJ ISSN: 0884-0431
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 1993:23157799 BIOTECHNO
AB Determination of the serum concentration of the protein **osteocalcin** (OC) is useful for the noninvasive evaluation of bone metabolism. Because the dog is an excellent experimental model for the study of bone, we produced and characterized a polyclonal antiserum specific for dog OC and used it to develop a radioimmunoassay (RIA) for the measurement of the concentration of this protein in dog serum. The antiserum expresses higher affinity for **Ca**.sup.2.sup.+-bound than for **Ca**.sup.2.sup.+-free OC (B.sub.5.sub.0 at 10.sup.-.sup.5 versus 2 x 10.sup.-.sup.4 dilution). Also, in the presence of **Ca**.sup.2.sup.+ affinity is higher for the carboxylated than for the decarboxylated form of the protein, and under **Ca** .sup.2.sup.+-free conditions the affinity is equal for the two forms. The study of peptide fragments of OC demonstrates competitive binding of the peptide comprising amino acids 20-44 but not of other fragments; this suggests that the antigenic epitope of dog OC is located in the midmolecular region of the protein. The RIA displays excellent sensitivity for the measurement of OC in blood (detection limit 0.31 ng/ml), with intraassay and interassay variations of 4.6 and 6.8%, respectively. Analysis of gel chromatography fractions of normal dog serum shows that greater than 90% of the antigenic material coelutes with purified radiolabeled dog OC. Test of parallelism reveals lack of interference of serum constituents with the binding assay. The antiserum displays limited species specificity since it cross-reacts with human OC, but not with the protein from rodents. Consistent with previous observations in other in vivo models, the serum concentration of OC in experimental dogs is decreased significantly 7-10 days after thyroparathyroidectomy and it is unchanged 1 month following ovariohysterectomy.

L10 ANSWER 10 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
ACCESSION NUMBER: 1988:18262661 BIOTECHNO
TITLE: Specific radioimmunoassay for ovine bone gla-protein (**osteocalcin**)
AUTHOR: Pastoureau P.; Merle B.; Delmas P.D.
CORPORATE SOURCE: Unite INSERM 234, Hopital E. Herriot, F-69437 Lyon, France.
SOURCE: Acta Endocrinologica, (1988), 119/1 (152-160)
CODEN: ACENA7 ISSN: 0001-5598
DOCUMENT TYPE: Journal; Article
COUNTRY: Denmark
LANGUAGE: English
SUMMARY LANGUAGE: English
AN 1988:18262661 BIOTECHNO
AB We developed a sensitive and specific radioimmunoassay for ovine bone gla-protein (**osteocalcin**) using a polyclonal rabbit **antibody** raised against ovine bone gla-protein. Bone from lambs was extracted in 0.5 mol/l **EDTA** and desalted on Sephadex G-25. Bone gla-protein was purified by gel filtration chromatography over Sephadex G-100 and ion-exchange chromatography on DEAE-Sephadex A-25. The protein, subjected to monodimensional electrophoresis migrated as a single spot in SDS PAGE with the same apparent molecular weight of 12 kD as bovine bone gla-protein. The amino acid composition of purified bone gla-protein was in agreement with a previous publication. The competitive RIA uses .sup.1.sup.2.sup.5I-labelled bone gla-protein as a tracer and a complex of a second **antibody** and polyethylene glycol to separate free and **antibody**-bound .sup.1.sup.2.sup.5I-labelled bone gla-protein. The intra- and inter-assay variations are less than 6 and 10%, respectively. There is no reactivity of our antisera with dog sera. The cross-reactivity is only partial with calf and human sera and complete with ovine sera. We measured bone gla-protein levels in serum of 96 normal male sheep of different ages. Serum bone gla-protein rapidly and significantly ($P < 0.001$) decreased from 535 ± 169 μg/l at birth, to 240 ± 43 μg/l at 45 days, 152 ± 44 μg/l at 90 days, and 5.9 ± 0.7 μg/l at 7 years of age. In addition, bone gla-protein levels at birth were higher in normal birth weight than in hypotrophic lambs with low birth weight (535 ± 169 vs 271 ± 156 μg/l, $P < 0.001$). Furthermore, lambs raised outside in free conditions tended to have higher serum bone gla-protein levels than lambs raised under shelter (194 ± 53 vs 137 ± 34 μg/l), suggesting a role of breeding factors such as diet or relative immunobilization on bone gla-protein levels. These results emphasize the interest of a RIA for the bone-specific protein gla-protein as a potential tool for experimental studies on skeletal growth and bone remodelling in a large animal.

L10 ANSWER 11 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
ACCESSION NUMBER: 1987:17113169 BIOTECHNO
TITLE: Expression of differentiated function by mineralizing cultures of chicken osteoblasts
AUTHOR: Gerstenfeld L.C.; Chipman S.D.; Glowacki J.; Lian J.B.
CORPORATE SOURCE: Laboratory for the Study of Skeletal Disorders and Rehabilitation, Children's Hospital, MA 02115, United States.
SOURCE: Developmental Biology, (1987), 122/1 (49-60)
CODEN: DEBIAO
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
AN 1987:17113169 BIOTECHNO
AB This report documents osteoblast differentiation in vitro, as demonstrated by the 50-100x increase of proteins which are known markers of the osteoblast phenotype. Collagen type I and **osteocalcin** synthesis and accumulation, alkaline phosphatase activity, and matrix

calcification show similar temporal relationships that are analogous to those seen during in vivo bone development. Chicken embryonic osteoblast progenitor cells were selected by initial growth at low densities in minimal medium. Upon subcultivation into nutrient-enriched medium at higher cell densities, near homogeneous populations of osteoblasts were obtained as demonstrated by the greater than 80% enrichment of cells positive for alkaline phosphatase activity. A comparison was made between cells grown in the presence or absence of 10 mM β-glycerolphosphate (β-GPO.sub.4), a chemical stimulant of matrix calcification, as a function of time. Cultures treated with β-GPO.sub.4 showed visible calcification at Day 12 when culture monolayers became confluent. By Day 30, numerous large foci of calcification were visible and a 20-fold increase in calcium (**Ca**) content was observed. In contrast, untreated cultures had only a 3-fold increase in **Ca** content with many smaller diffuse areas of calcification. DNA, RNA and total protein levels were nearly identical between the two cultures, indicating that β-GPO.sub.4 had no marked effect on either cell proliferation or transcriptional activity. The major collagen type produced by either culture was type I, with no detectable type III as determined by CNBr peptide mapping and delayed reduction analysis. Alkaline phosphatase activity showed a rapid .sim.50-fold induction by Day 18 and remained elevated in control cultures. However, cultures treated with β-GPO.sub.4 demonstrated a rapid 80% decline of enzyme activity after 18 days. In contrast, total **osteocalcin** levels showed a 100-fold induction by Day 18 remained elevated in both control and β-GPO.sub.4-treated cultures throughout the time period examined. While the overall levels of **osteocalcin** were the same in β-GPO.sub.4-treated and untreated cultures, 2- to 5-fold more **osteocalcin** was associated with the more mineralized matrices of the β-GPO.sub.4-treated cultures. In order to confirm the association of **osteocalcin** with areas of mineralization, co-localization of mineral to **osteocalcin** and collagen was carried out by combining vital labeling with tetracycline and immunofluorescent staining with anti-oseocalcin and anti-collagen **antibodies**. Both collagen and **osteocalcin** showed strong localization with areas of mineralization. This culture system defined the expression of specific markers of osteoblast function during differentiation and their relationships to matrix calcification. Thus, these osteoblast cultures provide a unique model in which to study the regulation of bone-specific proteins and genes during osteoblast differentiation and matrix calcification.

L10 ANSWER 12 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
ACCESSION NUMBER: 1986:16008948 BIOTECHNO
TITLE: Radioimmunoassay for human **osteocalcin** using an **antibody** raised against the synthetic human (h37-49) sequence
AUTHOR: Juppner H.; Schettler T.; Giebel G.; et al.
CORPORATE SOURCE: Kinderklinik, Abteilung fur Klinische Endokrinologie, Medizinische Hochschule Hannover, 3000 Hannover 61, Germany.
SOURCE: Calcified Tissue International, (1986), 39/5 (310-315) CODEN: CTINDZ
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
AN 1986:16008948 BIOTECHNO
AB Radioiodination of synthetic human 37-49 **osteocalcin** requires optimal labeling conditions in order to obtain a maximum of mono- and di-iodinated tracer with little contamination by tri- and tetra-iodinated products or 'radio-damage'. The **antibody** raised against **osteocalcin**(h37-49) had the highest affinity for the C-terminal peptide used for iodination and the larger peptide (h30-49). The intact bovine **osteocalcin** (b1-49) revealed less immunoreactivity. This

C-terminal specific radioimmunoassay detected the intact human **osteocalcin** in HPLC purified plasma and peritoneal dialysate from patients with terminal renal insufficiency and in extracted human bone. Some quantities of **osteocalcin** peptides with a higher hydrophobicity were predominantly detected in uremic plasma. These peptide that had a higher molecular weight than the intact human molecule might represent aggregated forms of the intact bone-derived **osteocalcin**. Immunoreactivity in plasma samples from healthy individuals revealed a remarkable difference as to which substance was employed for anticoagulation. Compared to heparin, the addition of **EDTA** largely reduced the **osteocalcin** immunoreactivity, implying that conformational changes within the N-terminal portion (containing the Gla- and Cys-residues) are extended to the C-terminal portion.

L10 ANSWER 13 OF 15 BIOTECHNO COPYRIGHT 2004 Elsevier Science B.V. on STN
ACCESSION NUMBER: 1986:16024766 BIOTECHNO
TITLE: Calcium-specific immunoassays for factor IX: Reduced levels of antigen in patients with vitamin K disorders
AUTHOR: Bray G.L.; Weinmann A.F.; Thompson A.R.
CORPORATE SOURCE: Division of Hematology, Department of Medicine, University of Washington School of Medicine, Seattle, WA, United States.
SOURCE: Journal of Laboratory and Clinical Medicine, (1986), 107/3 (269-278)
CODEN: JLCMAK
DOCUMENT TYPE: Journal; Article
COUNTRY: United States
LANGUAGE: English
AN 1986:16024766 BIOTECHNO
AB Polyclonal rabbit anti-factor IX antisera were fractionated to establish solid-phase immunoassays recognizing calcium-dependent and non-calcium-dependent epitopes. The assays were >99.9% specific for factor IX and sensitive to 0.05 U/dl plasma or 2 ng/ml purified factor IX. For the calcium-dependent fraction, an absolute requirement of divalent metal ions was found, and Sr(II), Mn(II), and Mg(II) could substitute for **Ca**(II). On immunoblots of reduced, electrophoresed factor IXa, the .sup.1.sup.2.sup.5I-calcium-dependent **antibody** fraction bound to the amino-terminal light chain. Plasma sampled from 13 patients receiving warfarin and one with cephalosporin-related vitamin K deficiency had a mean level of calcium-dependent factor IX antigen of 22 U/dl, comparable to the 24 jU/dl average of factor IX procoagulant activity; these two results were highly correlated. Antigen levels determined by either the polyclonal or a monoclonal, non-calcium-dependent anti-factor IX assays ranged from 1.7-fold to 6.0-fold greater than the corresponding levels of factor IX procoagulant activity or calcium-dependent antigen level for each subject's plasma. The difference reflects inactive, circulating factor IX. In contrast, factor IX antigen levels determined by an assay using a monoclonal, calcium-dependent anti-factor IX were from one half to one thirteenth as much as those measured by the polyclonal, calcium-dependent immunoassay. The disparity between results of calcium-dependent assays suggest that some Gla residues near the amino terminus of factor IX are relatively less important for normal procoagulant function of factor IX than others, are more sensitive to the effects of vitamin K antagonism or deficiency, and are important for the epitope recognized by this particular calcium-dependent, monoclonal **antibody**.

L10 ANSWER 14 OF 15 LIFESCI COPYRIGHT 2004 CSA on STN
ACCESSION NUMBER: 84:38059 LIFESCI
TITLE: Immunochemical studies of conformational alterations in bone gamma -carboxyglutamic acid containing protein.
AUTHOR: Delmas, P.D.; Stenner, D.D.; Romberg, R.W.; Riggs, B.L.; Mann, K.G.

CORPORATE SOURCE: Mayo Clin./Found., Rochester, MN 55905, USA
SOURCE: BIOCHEMISTRY (WASH.)., (1984) vol. 23, no. 20, pp. 4720-4725.
DOCUMENT TYPE: Journal
FILE SEGMENT: T; L
LANGUAGE: English
SUMMARY LANGUAGE: English

AB The **Ca** super(2+)-dependent transition of the vitamin K dependent bone protein bond Gla-containing protein (BGP) was investigated by use of anti-BGP **antibody** that reacts with the **Ca** super(2+)-dependent conformation of BGP. **Antibody** binding occurred in the presence of **Ca** super(2+) or Mg super(2+) with a K sub(d)(app) of 1.75 mM for **Ca** super(2+). Upon removal of **Ca** super(2+) with ethylenediaminetetraacetic acid, **antibody** binding was eliminated. Upon thermal acid decarboxylation of BGP, **Ca** super(2+)-independent binding of the **antibody** was restored. Thus, the epitope not expressed by fully carboxylated BFP in the absence of calcium ion was restored either by addition of **Ca** super(2+) or by decarboxylation of the protein.

L10 ANSWER 15 OF 15 CONFSCI COPYRIGHT 2004 CSA on STN
ACCESSION NUMBER: 91:24100 CONFSCI
DOCUMENT NUMBER: 91052897
TITLE: Selective **antibody** recognition of the **Ca** super(2+)-dependent conformation of bovine **osteocalcin**
AUTHOR: Neugebauer, B.M.; Gundlach, G.
CORPORATE SOURCE: Justus-Liebig-Univ., Giessen, FRG
SOURCE: FASEB, 9650 Rockville Pike, Bethesda, MD 20814, USA, Abstracts, FASEB Journal Poster Paper.
Meeting Info.: 912 0204: 75th Annual Meeting of FASEB (9120204). Atlanta, GA (USA). 21-25 Apr 1991. Federation of American Societies for Experimental Biology.
DOCUMENT TYPE: Conference
FILE SEGMENT: DCCP
LANGUAGE: UNAVAILABLE